Collagenase from Clostridium histolyticum (not sterile)

Cells of animal tissue are attached to each other by a complex matrix of proteins, glycoproteins, lipides, glycolipides, and mucopolysaccharides. To isolate individual cells or to start primary cultures, the delicate matrix must be partially broken down without destroying the cell or its surface. The treatment of tissue with Collagenase effects a careful, selective reduction of the intercellular matrix and does not influence the growth ability of the cells. The raw Collagenase offered by Biochrom AG is a mixture of different proteolytically efficient enzymes.

For optimal results, an exactly adjusted mixture of the proteolytic enzymes is necessary. For this purpose we offer 4 different types of Collagenase, CLS I, CLS II, CLS III, and CLS IV. The last one is normally applied with other enzymes like trypsin, elastase, or hyaluronidase. The Trypsin or Trypsin/EDTA generally used in cell cultures attacks the matrix only slowly, causing moreover irreversible damages of the released cells.

Product	Cat. No.	Unit
Collagenase type I, CLS I Enzyme blends of collagenase, clostripain, with tryptic and proteolytic activities from *Clostridium histolyticum*. This preparation is recommended for liver, lung, adipose, and adrenal tissue. Specific activity is 125 to 250 Mandl units per ml of powder substance. Storage temperature: +2 – +8 °C	C 1-28 C 1-22	100 mg 1 g
Collagenase type II, CLS II Enzyme blends from collagenase, clostripain, with tryptic and proteolytic activities, from *Clostridium histolyticum*. This preparation shows a high clostripain activity with the tryptic activity close to type I. Suitable for liver, bone, thyroid, heart, and salivary gland tissue. Specific activity is 125 to 250 Mandl units per milligram of powdered substance. Storage temperature: +2 – +8 °C	C 2-28 C 2-22	100 mg 1 g
Collagenase type III, CLS III Enzyme blends from collagenase, clostripain, with tryptic and proteolytic activities, from *Clostridium histolyticum*. This preparation has a very low proteolytic and a normal collagenase activity. Suitable for mammary glandular tissue. Specific activity is 125 to 250 Mandl units per milligram of powdered substance. Storage temperature: +2 – +8 °C	C 3-28 C 3-22	100 mg 1 g
Collagenase type IV, CLS IV Enzyme blends from collagenase, clostripain, with tryptic and proteolytic activities. Recommended for the isolation of Islets of Langerhans. Specific activity is 125 to 250 Mandl units per milligram of powdered substance. Storage temperature: +2 – +8 °C	C 4-28 C 4-22	100 mg 1 g

Recommendations

Actual enzyme concentrations should be 0.1 to 0.2 % (w/v), depending on the delicate nature of the tissue substrate. This is valid for an activity of about 160 Mandl units per milligram of dry substance. On other specific activities the concentration of use has to be modified correspondingly. The indication of the specific activity is related to the respective method of determination. The determination of activity according to Mandl is difficult to designate due to the high molecular collagen substrate. As it is a biological product, collagenase is subject to natural variations from charge to charge. Thus, Biochrom AG recommends to previously test deliverable charges concerning their suitability for the desired purpose and to optimize the process conditions (concentration, incubation time, and temperature).

- Definitions:
  Collagenase activity according to Mandl: 1 unit releases 1 µmol amino acids determined with ninhydrin and calculated as leucin from native collagen within 5 hours (+37 °C and pH 7.5).

- Collagenase activity according to Wünsch and Heidrich:
  1 PZ (4-phenyl-azobenzyl-oxycarbonyl) unit catalyses the hydrolysis of 1 µmol of water soluble chromo-peptide PZ-Pro-Leu-Gly-Pro-D-Arg per minute (+25 °C/pH 7.1). Resulting PZ-L-Pro-Leu can be determined.

In the table 44 on page 105 you will find the comparison of both enzymes Collagenase, and Trypsin concerning their properties.

Reference:

Tissue Culture, Methods and Applications (ed. P.F. Kruse and M.K. Patterson) Academic Press, New York [1972]